



# Marine Biological Research and Possible Application

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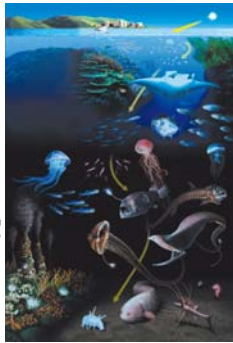
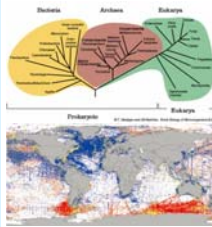
2 -3 May 2013, New York  
Intersessional Workshop on Marine Genetic Resources



## Exploring Marine Life

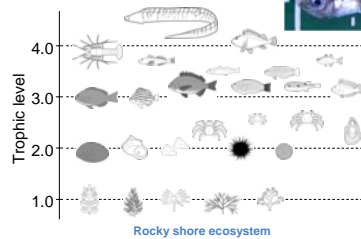
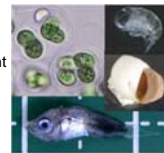
### Studies on the origin of life and the mechanisms of its evolution

- From the origin of life to early ecosystem evolution
- Evolution of eukaryotes
- Progress from monocellular to multicellular organisms



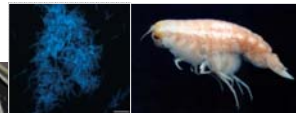
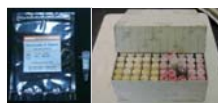
### Studies of the structures and functions of the Earth's biosphere, particularly in the ocean

- Biogeochemical cycles and their history
- Monitoring of the marine environment
- Adaptive ecology of organisms in extreme environments



### Applied research on microorganisms and functional molecules such as biological materials and enzymes









- Development of reagents and materials using useful enzymes and membrane substances



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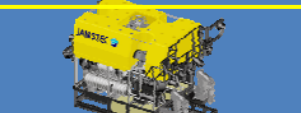
# Vessels

GT: Gross tonnage

<p><b>R/V NATSUSIMA</b> GT 1,739 t</p>  <p>Various observations with ROV</p>	<p><b>R/V KAIYO</b> GT 3,350 t</p>  <p>SWATH-type vessel with large workspace</p>	<p><b>R/V HAKUHOMARU</b> GT 3,991 t</p>  <p>Multipurpose research vessel with long-term cruise</p>
<p><b>R/V YOKOSUKA</b> GT 4,439 t</p>  <p>Support vessel for "SHINKAI6500"</p>	<p><b>R/V MIRAI</b> GT 8,687 t</p>  <p>Large vessel able to perform observation over wide areas</p>	<p><b>R/V KAIREI</b> GT 4,517 t</p>  <p>Surveys the structure of sub-bottoms mainly with MCS</p>
<p><b>D/V CHIKYU</b> GT 56,752 t</p>  <p>Drilling vessel with world-class scientific drilling capacity</p>	<p><b>R/V SHINSEIMARU</b> GT 1,600 t (approx.)</p>  <p>Operation from 2013 Multipurpose R/V with an emphasis on survey off the coast of TOHOKU</p>	<p><b>R/V (under planning)</b> GT 5,000 t (approx.)</p>  <p>Operation from 2015 Survey for submarine resources</p>

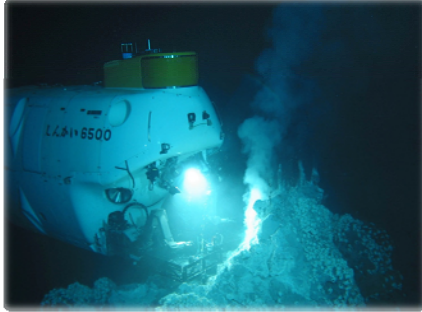
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# Manned/Unmanned Underwater Vehicles

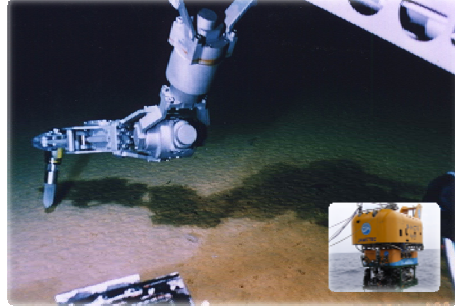
<p>Deep Submergence Vehicle</p> <p><b>Shinkai6500</b></p>  <p>World-class manned submersible</p>	<p><b>Shinkai2000</b> (Retired)</p>  <p>Japan's first full-fledged manned submersible for deep-sea research</p>	<p><b>URASHIMA</b></p>  <p>Large AUV capable of long-distance dives</p>
<p><b>ABISMO</b></p>  <p>Automatic Bottom Inspection and Sampling Mobile</p>	<p><b>AUTONAVPICASSO</b></p>  <p>Plankton Investigatory Collaborating Autonomous Survey System Operon</p>	
<p><b>HYPER-DOLPHIN</b></p>  <p>Operation with high-sensitivity camera and manipulators</p>	<p><b>KAIKO 7000 II</b></p>  <p>Capable of diving up to a maximum depth of 7,000 m</p>	<p><b>ROV under construction</b></p>  <p>ROV for heavy-duty work To be launched in 2013</p>



## Examples of Sampling



**Manned Research Submersible**



**ROV**

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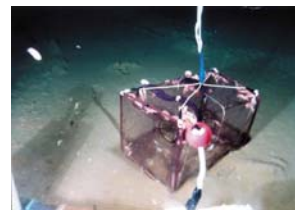
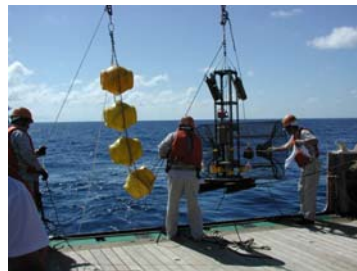
## Examples of Sampling



**Multiple Corer**



**Piston Corer**



**Bait Trap**

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## Sample Analysis Laboratory

### The state-of-the-art facilities for Life and Earth Planetary Sciences

Toward systematic understanding of **Life, Ocean and our planet Earth** through core sample research, Kochi Institute for Core Sample Research will promote the overarching approaches using **ultra-high resolution mass and molecular analyses**.



Secondary ion mass spectrometry (NanoSIMS 50L and IMS1280HR)



Single Cell Analysis Lab. in ultra-clean room



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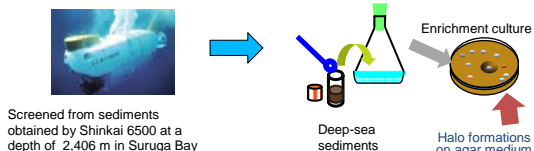
## Development of findings ( Nucleic acid extraction from Agarose gel )

Relatively large nucleotide fragments can be recovered without damage so that quick DNA and RNA purification became possible.

### (1) Screening of agarase-producing microorganisms from deep-sea sediments

- AGAR-DIGESTING ENZYME AND UTILIZATION THEREOF (Japanese Patent No. 4441486)

Screening of agarase-producing microorganisms from deep-sea sediments



Licensed

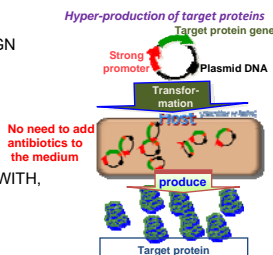
**NIPPON GENE**



### (2) Hyper-production of target proteins

- METHOD FOR STABLY RETAINING FOREIGN GENES IN CELLS (Japanese Unexamined Patent Application Publication Number 2009-17840)

- NOVEL DNA FRAGMENT, RECOMBINANT VECTOR COMPRISING SAME, TRANSFORMANT TRANSFORMED THEREWITH, AND USE THEREOF (Japanese patent No. 5126879)



Thermostable  $\beta$ -Agarase

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## What is *Hirondellea gigas*?



- They live in the Challenger deep (the deepest point in the world).
- They belong to Amphipoda.
- They were captured during a R/V cruise by a bait trap with a cuttlefish (Miyake et al., 2009).



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## Mysteries of *Hirondellea gigas*



- What does *Hirondellea gigas* eat?
- ✓ Very low population of fish/plankton in the deep sea
- Microorganisms?
- ✓ Very rare in deep-sea sediment
- Digestive enzymes?

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## Digestive enzymes

Enzyme detection using mashed *H. gigas*

Detected enzyme activities	Substrate	Source
• Protease	→ Protein	} Animals
• Lipase	→ Lipid	
• Amylase	→ Starch, glycogen	} Plants
• Cellulase	→ Cellulose	
• Glucomannanase	→ Glucomannan	
• Maltase	→ Maltose	

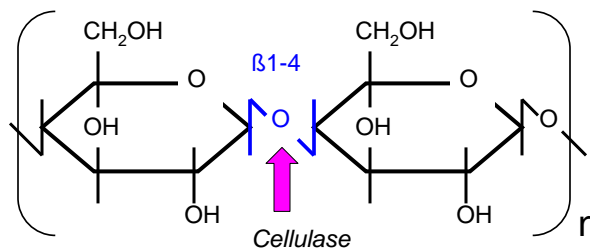
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## Cellulase degrades cellulose

### What is cellulose?

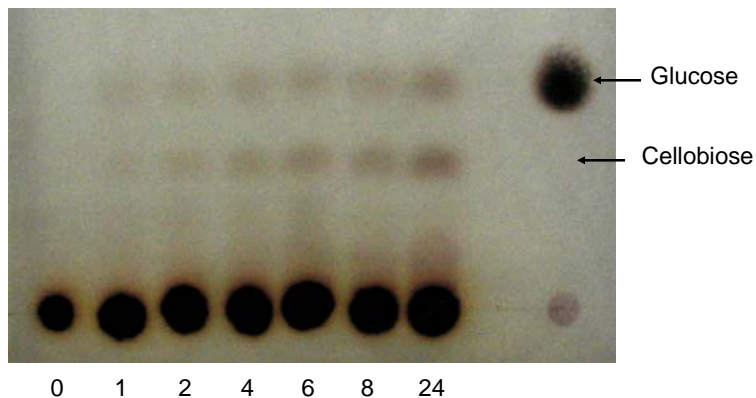
- Cellulose is mainly contained in the green plants, algae and oomycetes.
- Cellulose is the most common organic carbon on Earth.
- Cellulose is a polymer of glucose ( $\beta$ 1-4).



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## TLC analysis of products



0 1 2 4 6 8 24

Reaction time (h)

30°C, 10 mM Sodium acetate (pH 5.6)

Substrate; 1% Carboxymethyl-cellulose (CMC)

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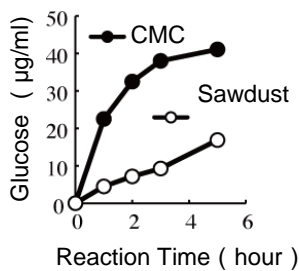
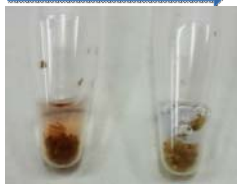


## Reaction with Sawdust and Recycled Paper

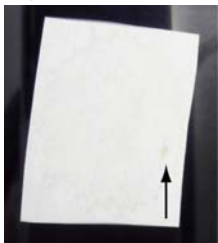
### Sawdust (Oak)



Enzyme reaction  
at 35°C



### Recycled Paper



Enzyme reaction  
at room temperature



Glucose in a  
red colour

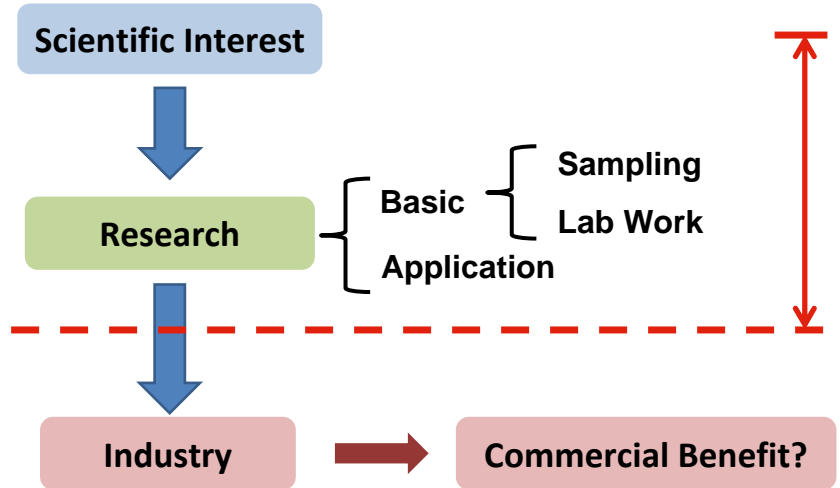
Before enzyme reaction

15 hours later

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## What drives scientists?



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### Acknowledgement:

Special thanks to  
Dr Hideki Kobayashi, JAMSTEC  
Dr Yuji Hatada, JAMSTEC

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